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Use of different leguminous seeds for lactating goats. Amino acid composition of the raw material and the rumen undegradable fraction

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SUMMARY – The suitability of different leguminous seeds (lupine, beans, bitter vetch and vetch) for the nutrition of goats was investigated. The ruminal degradability of the N in each protein source was estimated using the nylon bag technique. The amino acid composition of the original protein sources and of the residues in the bags after ruminal incubation was also determined. The beans had more ($P<0.05$) of the quickly degradable fraction of crude protein than did the other three leguminous seeds. Lupine seeds had a higher ($P<0.05$) rate of degradation than did the other protein sources. The lowest value for effective degradability was observed for bitter vetch. The overall change in the amino acid profile due to ruminal fermentation depended on the protein source provided. The amino acid profile of the different leguminous seeds was among them, more similar before ruminal fermentation. However, the profile of their rumen undegradable protein fractions resulted very different. When a multivariate analysis was carried out, organization of the leguminous seeds in the order of the intensity of change, yielded the following: lupine, beans, bitter vetch and vetch.

Keywords: Leguminous seeds, raw material, rumen undegradable protein, amino acid profile, goats.

RESUME *– "Utilisation de graines de différentes légumineuses dans les régimes pour chèvres en lactation. Composition en acides aminés des matières premières et de la fraction indégradable dans le rumen". Dans cette étude a été analysée l'aptitude de l'usage de diverses légumineuses en graines (lupin, fève, ers et vesce), dans l'alimentation de la chèvre. La dégradabilité du N de chacune des sources protéiques a été réalisée au moyen de la technique du sachet en nylon. De la même façon on a déterminé la composition en acides aminés des sources originales de protéines et des résidus contenus dans le sachet. Par rapport aux trois autres légumineuses, les fèves ont présenté une fraction supérieure ($P<0,05$) de protéines rapidement dégradables. Le lupin, lui, face aux trois autres légumineuses, a présenté un taux de dégradation plus élevé ($P<0,05$). Les valeurs les plus faibles de dégradation effective ont correspondu aux ers. Le changement subi dans la composition en acides aminés dû à la fermentation de la rumination, dépendait de la source protéique. Le profil amino-acidique des différentes graines de légumineuses se trouvait être semblable avant la fermentation de la rumination. Au contraire, le profil correspondant aux fractions non dégradables par rumination, se trouvait être relativement différent. En réalisant une analyse factorielle à multiples variantes, les différentes légumineuses se sont trouvées classées selon l'intensité du changement subi dans leur composition en acides aminés, de la manière suivante : lupin, fève, ers et vesce.*

Mots-clés : *Graines de légumineuses, matières premières, protéine non dégradable lors de la rumination, profil amino-acidique, chèvres.*

Introduction

The suitability of a protein source for the nutrition of goats or other ruminant species depends on the fraction of this protein source that after ruminal degradation gives rise to microbial protein, and to the fraction that escapes ruminal fermentation and by reason of its aminoacid composition, may supplement microbial protein in the intestine. Therefore, the goal of the present study was to determine the aminoacid composition of four leguminous seeds (lupine, beans, bitter vetch and vetch), and to identify the changes in such composition that occur after these leguminous seeds are transformed in the rumen.

Material and methods

Degradability and aminoacid composition of the different protein sources

The ruminal degradability of the N in each protein source was estimated using the nylon bag technique according to an international standard procedure (Madsen and Hvelplund, 1994). Three goats with permanent ruminal cannulas were used for incubations of 0, 4, 8, 16, 24 and 48 h. The diet fed to the cannulated goats was that proposed by ∅rskov and McDonald (1979). The diet consisted of a good quality alfalfa hay with a mineral and vitamin supplement. For each incubation time, two bags were used per animal. The dimensions of the bags were 7.5 x 10 cm, and the pore size was 46x46 μm. The amount of sample per bag was 2 to 3 g, which were previously milled through a 2.0 mm screen. After incubation, the bags were washed in a washing machine and were treated in a stomacher for 4 min to remove microbial contamination (Madsen and Hvelplund, 1994).

The aminoacid composition of the original protein sources and of the residues in the bags after ruminal incubation was determined by HPLC using the Waters Pico-Tag method, which involves precolumn derivatization with phenylisothiocyanate. Protein hydrolysis was performed in 6 M HCl in sealed, evacuated tubes at 110ºC for 24 h.

Statistical analysis

The degradation kinetic parameters of each protein source were estimated using the model of ∅rskov and McDonald (1979). The effective degradability was calculated as a + (b x c/c + k) where a = quickly degradable fraction, b= slowly degradable fraction, c = rate of degradation and k = rate of passage of the ingesta. The k value used was equal to 0.031/h (Molina *et al*., 2000). The obtained results were analysed statistically in accordance with the general linear model procedure as described by Steel and Torrie (1984). The model accounts for variations caused by the protein source. Statistical analyses were performed using the Statgraphics Statistical package (Statgraphics, 1991).

With the aim to establish the overall change in the aminoacid composition of protein sources after ruminal incubation, a multivariate analysis was performed using the factor procedure of SAS (1987), the algorithm used was PROC FACTOR. The correlation matrix was selected. The methods for the factors extraction and rotation were principal components analysis and varimax, respectively (SAS, 1987). The number of factors derived in each case depended on the fraction of the total variance that each of the factors explained as well as on the extent to which each variable defined them. The experimental units were the protein sources both before and after ruminal fermentation. The variables considered were the concentrations of each of the essential and nonessential amino acids.

Results and discussion

Degradability of the protein sources

The degradation characteristics of the crude protein of the protein sources in the rumen are shown in Table 1. The beans had more ($P<0.05$) of the quickly degradable protein of crude protein than did the other three leguminous seeds. In general, the concentration of slowly degradable fractions of crude protein were greater ($P<0.05$) than for the quickly degradable fractions. Lupine seeds had a higher ($P<0.05$) rate of degradation than did the other protein sources. The lowest values for effective degradability were observed for bitter vetch.

Of the protein sources here considered, it is well known that goats show a high preference for bean seeds. The other three sources were chosen because they may be considered as an appropriated alternative source of meat meals for animal feeding.

Aminoacid composition of protein sources and of rumen undegradable protein (RUP) fractions of protein sources

Table 2 shows the aminoacid composition of the protein sources and residues in the bags after 48 h of incubation. The different leguminous seeds had similar total essential aminoacid content. These values were similar before and after ruminal incubation. Due to the ruminal fermentation, there was a marked decrease in the concentration of Arg in the protein sources, except in vetch seeds. The concentration of His increased in all the protein sources while that of Lys decreased in the case of the beans and vetch seeds, remained unchanged in the case of bitter vetch seeds and increased considerably in the lupine seeds.

Table 1. Ruminal degradation parameters of the leguminous seeds[†]

	a		b		c		ED	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Lupine	43.41^{b}	0.36	56.54^{b,c}	0.79	14.21^{a}	8.03	88.48^{a}	3.56
Beans	54.44^{a}	0.73	51.65^{c}	4.86	5.55^{b}	2.65	85.91^{a,b}	2.75
Bitter vetch	43.58^{b}	1.30	62.29^{a,b}	7.70	5.27^{b}	1.67	81.80^{b,c}	2.45
Vetch	38.82^{c}	2.36	67.27^{a}	4.50	5.15^{b}	0.81	80.55^{c}	0.80

[†]a=Quickly degradable fraction, b=slowly degradable fraction, c=rate of degradation and ED=effective degradability; ^{a,b,c} $P<0.05$.

Table 2. Aminoacid composition (g/100g AA) of the leguminous seeds and the residues in the bag after 48 h of ruminal incubation

	Lupine		Beans		Bitter vetch		Vetch	
	Feed	Residue	Feed	Residue	Feed	Residue	Feed	Residue
EAA								
Lys	4.97	7.99	6.97	5.78	6.71	6.81	7.24	4.84
His	2.60	2.90	2.30	4.47	2.50	3.39	2.46	2.95
Thr	3.42	4.83	3.71	3.59	3.64	2.25	3.55	3.17
Arg	11.56	3.60	10.24	7.46	9.06	7.30	7.63	7.25
Val	4.33	7.22	5.49	6.63	5.16	5.76	5.59	6.20
Ile	4.18	4.54	4.27	3.95	4.07	4.22	4.52	4.47
Leu	6.99	7.36	7.92	6.47	7.40	8.24	7.33	6.89
Phe	4.06	5.94	4.64	4.39	4.29	4.79	5.08	4.82
Total	42.11	44.38	45.54	42.74	42.83	42.76	43.40	40.59
NEAA								
Tyr	4.15	4.26	3.47	7.04	3.38	4.73	3.81	4.47
Asp	11.25	13.76	12.11	8.42	13.65	11.72	13.08	12.35
Glu	23.14	9.31	17.24	9.59	18.90	12.91	18.25	15.35
Ser	5.65	9.70	5.57	6.36	5.47	6.56	5.65	6.40
Gly	4.56	5.43	4.74	14.09	4.76	8.49	4.95	7.72
Ala	4.18	5.85	5.16	5.67	4.93	6.44	5.34	6.30
Pro	4.96	7.31	6.21	6.09	6.08	6.39	5.52	6.83
Total	57.89	55.62	54.50	57.26	57.17	57.24	56.60	59.42

EAA: Essential AA; NEAA: Nonessential AA.

To compare the overall changes in the aminoacid composition of each of the leguminous seeds that were caused by the action of the rumen, a multivariate analysis was carried out. The results of this are shown in Table 3 and Fig. 1. Two different factors were derived, the first and second accounting for 24.58 and 21.84% of the total variance, respectively. In accordance with the factor

loadings, the first factor was mainly determined by the concentration of Ser, Val, Phe and Pro with positive loading values and by the concentration of Arg and Glu with negative loading values. The second factor was mainly determined by the concentration of Gly, His and Tyr with positive loading values and by the concentration of Asp and Ile with negative loading values. In Fig. 1, the position of the different leguminous seeds before and after ruminal degradation and the situation of the different amino acids are represented in relation to these factors.

Table 3. Factor pattern for amino acids from leguminous seeds and residues in the bag after 48 h of ruminal incubation[†]

	Factor matrix	
Variable	Factor 1 [††]	Factor 2[†††]
Ser	0.9276	-0.0715
Val	0.9063	0.3580
Phe	0.8980	-0.2742
Pro	0.8466	0.0668
Ala	0.6398	0.3888
Thr	0.5027	-0.3378
Glu	-0.8054	-0.5697
Arg	-0.9788	-0.0813
Gly	0.1653	0.9663
His	0.2548	0.9251
Tyr	0.1891	0.9145
Ile	0.5138	-0.6091
Asp	0.2890	-0.8870
Leu	-0.0252	-0.3049
Lys	0.5179	-0.2727
	Final statistics	
Factor	Variance explained (%)	Cumulative variance explained (%)
1	24.58	24.58
2	21.84	46.42

[†]Results were derived from multivariate analysis.
[††]Express mainly by the Ser, Val, Phe, Pro, Arg and Glu content in the protein sources and residues in the bag.
[†††]Express mainly by the Gly, His, Tyr and Asp content in the protein sources and residues in the bag.

After examination of the effect of ruminal incubation in the aminoacid profile by multivariate analysis, some authors (Rulquim and Verité, 1993) reported that the changes detected were small in comparison with those that continue existing among feeds. However, other researchers (Erasmus *et al*., 1993; Cozzi *et al*., 1994; Sanz Sampelayo *et al*., 1999) reported data from which it can be deduced that the use of the original aminoacid profile of one protein source to predict amino acids available for absorption is not accurate because the change that is undergone differs according to the protein sources, such as was inferred from this study (Fig. 1). The aminoacid profile of the different protein sources was among them more similar before ruminal fermentation. However, the profile of their corresponding rumen undegradable fractions resulted very different. The degree of modification brought about by ruminal fermentation of the aminoacid profile of each protein source is given by the magnitude of the vector that joins the position of each one before and after ruminal fermentation. Two types of change in the aminoacid composition of the protein sources were observed: one in the positive direction of axis 1 (lupine) and the other in the positive direction of axis 2 (vetch, bitter vetch and beans). Organization of the feeds in the order of the intensity of change yielded the following: lupine, beans, bitter vetch and vetch.

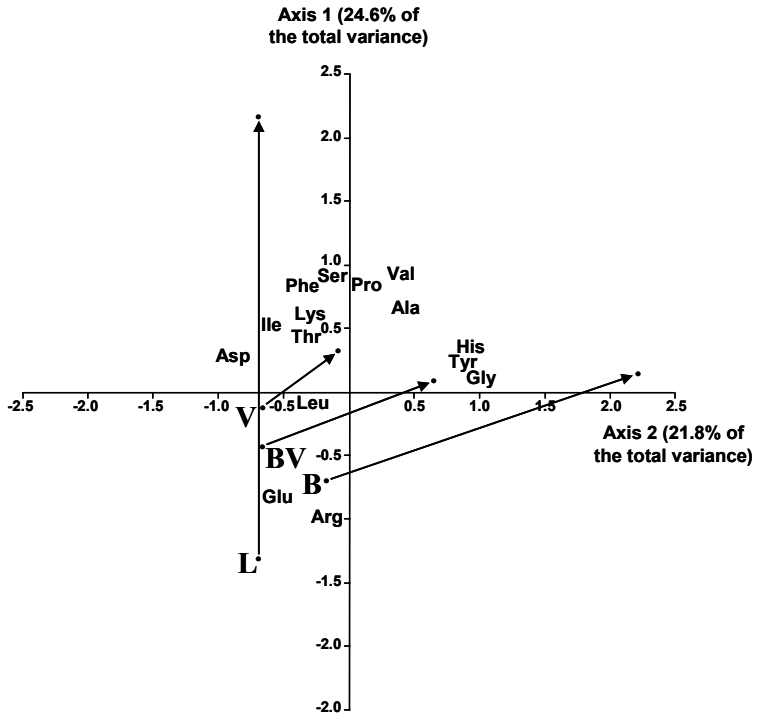

Fig. 1. Effects of ruminal degradation on the factor pattern of the aminoacid profile of the protein sources. L = Lupine; B = Beans; BV = Bitter vetch; V = Vetch. Point at end of arrow represents the factor pattern of the aminoacid profile of residue remaining in the bag.

Conclusion

In the goat and due to the ruminal fermentation, it is deduced that the overall change in the aminoacid profile of the different leguminous seeds here considered, depended on the protein sources in question. This aminoacid profile was between the different protein sources more similar before than after ruminal fermentation. When a multivariate analysis was carried out, organization of the leguminous seeds in the order of the intensity of change yielded the following: lupine, beans, bitter vetch and vetch.

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